

DOCKET NO: UPN0015-103/N2492
PATENT APPLICATION

Serial No.: 09/977,716
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IN THE CLAIMS:

This listing of claims will replace all prior versions, and listings, of claims in the application.

1. (Currently Amended) A method for detecting molecules expressing a selected epitope in a sample comprising:

(a) immobilizing a molecule expressing a selected epitope in a sample to a selected surface;

(b) contacting the surface with an epitope detector so that the epitope detector binds to immobilized molecules on the surface, said epitope detector comprising an oligonucleotide attached to a monoclonal antibody for the selected epitope, a single chain Fv for the epitope, ~~or~~ a constrained epitope specific CDR, a CDR mimetic or an engineered CDR structure, wherein said oligonucleotide

i) comprises an RNA promoter, and

ii) is attached to said monoclonal antibody, single chain Fv, constrained epitope specific CDR, CDR mimetic or engineered CDR structure by biotin-streptavidin linkers;

(c) amplifying the oligonucleotide of said epitope detector by RNA amplification to produce an amplified RNA product;

(d) contacting the amplified RNA oligonucleotide product with a fluorescent dye which binds to RNA and stains the amplified RNA oligonucleotide product; and

(e) detecting fluorescence emitted from the stained amplified RNA oligonucleotide product of step (d) which is indicative of epitope detector bound to the surface and molecules expressing the selected epitope in the sample.

2-14. (Canceled)

15. (Currently Amended) The method of claim 1 wherein the selected surface comprises an epitope anchor to immobilize the molecule expressing a selected epitope in

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~~a sample to the selected surface, to which the molecule expressing a selected epitope in a sample is immobilized is a chip or plastic well.~~

16. (Previously presented) The method of claim 1 wherein the selected surface is a chip or a microtiter plate.

17. (Canceled)

18. (Previously presented) The method of claim 1 wherein the oligonucleotide is a double stranded cDNA molecule.

19. (Previously presented) The method of claim 1 wherein the oligonucleotide comprises an RNA promoter selected from the group consisting of a T7 RNA promoter, a T3 RNA promoter, and an SP6 RNA promoter.

20. (Previously presented) The method of claim 1 wherein the fluorescent dye is an unsymmetrical cyanine dye.

21. (Currently Amended) The method of claim 1 ~~17~~, wherein said biotin is located at the 5'-terminus of the oligonucleotide.

22. (Currently Amended) The method of claim 1 further comprising adding the amplified RNA product ~~oligonucleotide~~ of said epitope detector from step (c) to a reverse transcriptase based reaction or a replicase based reaction to increase sensitivity.

23. (Currently Amended) A method for quantifying molecules expressing a selected epitope in a sample comprising:

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- (a) immobilizing a molecule expressing a selected epitope in a sample to a selected surface;
- (b) contacting the surface with an epitope detector so that the epitope detector binds to immobilized molecules on the surface, wherein said epitope detector comprises comprising an oligonucleotide attached to a monoclonal antibody for the selected epitope, a single chain Fv for the epitope, or a constrained epitope specific CDR, a CDR mimetic, or an engineered CDR, wherein said oligonucleotide comprises an RNA promoter;
- (c) amplifying the oligonucleotide of said epitope detector by RNA amplification to produce an amplified RNA product;
- (d) contacting the amplified RNA product oligonucleotide with a fluorescent dye which binds to RNA and stains the amplified RNA product oligonucleotide; and
- (e) measuring a quanta of fluorescence signals emitted from the stained amplified RNA product oligonucleotide which is directly proportional to epitope detector bound to the surface and molecules expressing the selected epitope in the sample.

24. (Currently Amended) The method of claim 23 wherein the selected surface comprises an epitope anchor to immobilize the molecule expressing a selected epitope in a sample to the selected surface, to which the molecule expressing a selected epitope in a sample is immobilized is a chip or plastic well.

25. (Previously presented) The method of claim 23 wherein the selected surface is a chip or a microtiter plate.

26. (Currently Amended) The method of claim 23 wherein wherein the oligonucleotide is linked to the monoclonal antibody, single chain Fv, constrained epitope specific CDR, CDR mimetic or engineered CDR structure by biotin-streptavidin linkers.

27. (Previously presented) The method of claim 23 wherein the

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oligonucleotide is a double stranded cDNA molecule.

28. (Previously presented) The method of claim 23 wherein the oligonucleotide comprises an RNA promoter selected from the group consisting of a T7 RNA promoter, a T3 RNA promoter, and an SP6 RNA promoter.
29. (Previously presented) The method of claim 23 wherein the fluorescent dye is an unsymmetrical cyanine dye.
30. (New) The method of claim 26 wherein said biotin is located at the 5'-terminus of the oligonucleotide.
31. (New) The method of claim 1 wherein said epitope detector comprises an oligonucleotide attached to a monoclonal antibody for the selected epitope.
32. (New) The method of claim 1 wherein said epitope detector comprises an oligonucleotide attached to a monoclonal antibody for the selected epitope.
33. (New) The method of claim 32 wherein the oligonucleotide is linked to the monoclonal antibody by biotin-streptavidin linkers.